Exhibit K-4
Appl. No. 09/558,421

pMC9 / Taq + D.V

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pool : Taq 10 + Deep Vent different amount in PCR ing
pMC9.

Deep Vent buffer   22X   Klentaq buffer.

	10x buffer	110		110		200 µM dNTP
dNTP		22		22		1 µM primer
Mg		1		22		200 pg Template
primer	1	11		11		Mg 2 mM
	2	11		11		
Template		4.4		4.4		
H2O		895.6		873.6		

added 10 Taq in 1 µl in 1x buffer

added different amount of Deep Vent in 2 µl in 1x buffer
either D.V. or R.T buffer

* Tubes.

Taq	Deep Vent		Deep Vent buffer		R.T. buffer	
0	0		1	2	22	23
1	0		3	4	24	25
	.001		5	6	26	27
	.005		7	8	28	29
	.01		9	10	30	31
	.05		11	12	32	33
	.1		13	14	34	35
	.5		15	16	36	37
	1		17	18	38	39
	2		19	20	40	41
			21		42	

94°, 3'
30 (94°, 30", 56°, 30", 72°, 3')   1 : 0.01   mix

started the cycling file instead of step file — 2 cycles done
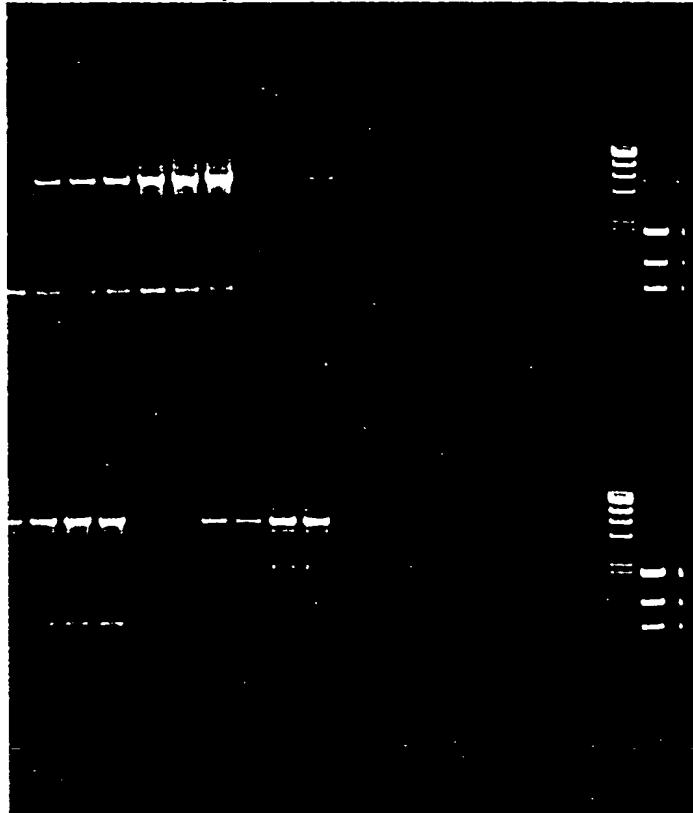before changing to step file .   To Page No._____

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Tag : Deepvent
titration

← D.V buffer — upto
1 : 0.01 ok.

← R.T. Buffer can go
up to 1: 0.05 but
less conc of D.V more
mispriming

maybe didn't add any Taq — make premix so next
- mispriming still there — check annealing temp.
- Try again with new D.vent.
Increasing D.V over 0.05 = having D.V alone.
Taq effect nil.

discarded 12/19/94

Witness d & Understood by me,
[signature]

Dat 11/28/94

Invented by

Recorded by R. Subramanian

Date 11/22/94